Editorial

DNA-Barcoding – a new tool to look at the diversity of life

Counting, classification and naming come naturally to human beings. From time immemorial, human beings have been naming and classifying life forms based on existing knowledge and the culture of the region. Modern scientific classification of species started with the development of binomial nomenclature by the Swedish botanist Carolus Linnaeus (1707-78). Till now, approximately 1.7 million species of plants and animals have been named (excluding microbes), and it is said that there could be 10 million species in the world. Thousands of new species are described every year, particularly invertebrates. Even in the well-known vertebrate groups, such as birds and mammals, new species are being discovered with increasing frequency, mainly in the tropics.

Earlier, species were mainly described based on their morphological features, including skeletons. In recent time, a new method – DNA barcoding or DNA taxonomy – has been developed to identify species based on their DNA sequence. In brief, the barcode of life is a short DNA sequence, from a uniform locality on the genome, used for identifying species. The proponents of DNA barcoding claim that 'it will help people quickly and cheaply recognize known species and retrieve information about them, and will speed discovery of the millions of species yet to be named'. They have given ten main reasons for Barcoding Life (http://phe.rockefeller.edu/barcode/docs/ TenReasonsBarcoding/pdf). We list a few of them here. Barcoding can identify a species from bits and pieces, such as morphologically unrecognizable products derived from protected species or tissue pieces of bird strikes to aircrafts. Even plant material in processed foodstuffs can be identified, which will greatly help in the quality control of foodstuff and traditional medicine. The second important reason and a great improvement of the traditional taxonomic approach is that Barcoding can identify a species in all life stages, from eggs, larvae, seed, pupa, adult, flower, leaf, root etc. This will help in controlling trade of protected species and the products derived from their various life stages, which are sometimes difficult to identify. Barcoding also allows relatively rapid identification of candidate species, which may turn out to be a new species, on which necessary morphological and taxonomic research can be focused. DNA barcoding can also unmask look-alikes. Many harmful insects masquerade as harmless ones, so once we identify the cheaters, necessary initiatives can be taken to control them. Barcoding will also help us preparing the 'life-trees' – the phylogenetic similarities, differences and evolutionary relatedness among taxa.

Before we go ahead and start analyzing specimens, we have to prove that DNA barcoding is effective in distinguishing between intraspecific and interspecific mtDNA variation. Fortunately, some recent studies are proving that barcodes can distinguish more than 95% of species (Ward *et al.* 2005; Hajibabaei *et al.* 2006). In an often-quoted paper, Hebert *et al.* (2004) have shown that in the 260 North American bird species, mitochondrial gene Cytochrome C Oxidase 1 (COI) variation between species was much greater than that within species. They found that differences between closely-related species were, on an average, 18 times higher than the differences within species. They also found that most of the species did not share the same barcode from those of any other species. Interestingly, their research also led to identification of cryptic species, which were earlier considered to be single species. This was further confirmed when their morphology and songs were analyzed. In 17 sets of species with overlapping barcode (Kerr *et al.* 2007), it was found that it could be due to three reasons: some may be recently diverged sister taxa where COI has not accumulated sequence differences, secondly, these species could be sharing mtDNA due to hybridization, and thirdly some of the species showing overlapping may be single species.

Kerr *et al.* (2007) have shown that "most provisional species were small to medium-sized, plainly coloured birds, whereas most species with overlapping barcodes were large and/or brightly coloured, which might reflect a natural taxonomic tendency towards under-splitting inconspicuous birds and/or over-splitting more conspicuous species." This is amply proved by the study of phylogeny of all species and nearly all subspecies of *Seicercus* and representatives of all subgenera in the *Phylloscopus* species of warblers (Olsson *et al.* 2004) and *Acrocephalus* genus (Leisler *et al.* 1997).

Recent DNA studies and genetics have confirmed the unity of the human being as single species, *Homo* sapiens. Comparison of COI barcode sequences shows that we differ from one another by only one or two base pairs out of 648, while we differ from the Chimpanzee (our closest relatives) at about 69 locations and the Gorilla at about 70 locations. Barcoding studies have also proved that there are two species of the Orangutan, and not one.

Bird taxonomy of the Indian subcontinent is going through radical changes, not only in nomenclature, but also in classification (e.g. Grimmett *et al.* 1998) and taxonomic upgradation of subspecies to full species or splits (e.g. Rasmussen and Anderton 2005). Rasmussen and Anderton (2005) have made 198 species-level changes in South Asia, which include many species splits within the region, splits within extra-limital species, and relocation of the race. For example, about 218 endemic bird species have been recorded from South Asia. In India, 18 subspecies have been upgraded to full species level, bringing the total to 79 fully endemic birds in India. The taxonomic status of some endemic species is not yet clear, and there is dispute over the status of some subspecies or races. Perhaps, DNA barcoding would help in settling such taxonomic uncertainties.

The Indian Council of Agricultural Research, New Delhi has asked the National Bureau of Fish Genetic Resources (NBFGR) to undertake an ambitious project to develop DNA barcoding of the Indian fish species. The scientists of NBFGR have already developed barcodes for about 50 fish species. This project is expected to go a long way for identification of species, subspecies and populations of our fish resources, which will also help in sustainable utilization, management and conservation.

There is a Consortium for the Barcode of Life (CBOL) whose aim is to have an international collaboration of natural history museums, herbaria, biological repositories and biodiversity inventory sites, together with academic and commercial experts in genomics, taxonomy, electronics, and computer sciences to speed up the compilation of DNA barcodes of all life forms. It also aims to establish a public library of sequences linked to named specimens, and promote development of portable devices for DNA barcoding. For more information visit: http://barcoding.si.ed. and http://www.barcodinglife.org

Until now only two studies have been carried out using the mtDNA technique on Indian birds, e.g. Largebilled Reed Warbler Acrocephalus orinus (Bensch and Pearson 2002; Round et al. 2007), and a new species of Scimitar-Babbler from Myanmar (Rappole et al. 2005). Genetech Institute, Colombo, Sri Lanka is likely to take up the responsibility of DNA barcoding of Sri Lankan birds. In the current scenario of rapid habitat changes, declining avifauna of the Indian subcontinent, and taxonomic confusion of many species, there is an urgent need to take up DNA barcoding of the Indian avifauna for their long-term protection. According to Jathar and Rahmani (2006), out of the 79 endemic Indian birds species, three are Critically Endangered, one Endangered, 14 Vulnerable, three Data Deficient, 15 Near Threatened, 27 Least Concern, and 16 require revision of their conservation status. Some of the endemic Indian birds have very limited distribution. For example, the Nilgiri Blue Robin Myiomela major and the White-bellied Blue Robin Myiomela albifentris were earlier considered as subspecies of the White-bellied Shortwing Brachypterex major. Rasmussen and Anderton (2005), based on morphological and vocal differences, have treated both as full species, and have also placed them under *Myiomela* and not under *Brachypterex*. In this case, there is taxonomic shift and taxonomic upgradation of the species. This taxonomic upgradation is of great conservation concern, because both species are found in a tiny range in the southern Western Ghats – the White-bellied Blue Robin is confined to densely wooded streams and Shola forests from Palni to Ashambu Hills of Kerala and Tamil Nadu, south of the Palghat Gap, mostly above 1600 m, while the Nilgiri Blue Robin is a resident bird of the Nilgiri Hills and the nearby Bababudan and Bramhabiri Hills, north of the Palghat Gap. Both are considered Vulnerable by BirdLife International (2007). The Barn Owl Tyto alba is one of the most widespread birds in the world and it is fairly common, with no threat of extinction. BirdLife put it in the Least Concern category. It is widespread in India, including the Andaman and Nicobar islands. Earlier, the individuals found in the Andamans were considered to be a distinct subspecies Tyto alba deroepstorffi (Baker 1927, Ripley 1961, Ali and Ripley 1969). Inskipp et al. (1996), Grimmett et al. (1998) and Kazmierczak and van Perlo (2000) recognized two subspecies: stertens found in the whole Indian subcontinent, and *deroepstorffi* found in the Andaman Island. However, Rasmussen and Anderton (2005) have treated the subspecies of the Andaman as a full species, Tyto deroepstorffi, based on the distinct morphological differences described by König et al. (1999). If DNA barcoding further corroborate that it is a full species, it means that we have to re-evaluate its conservation status. Similar is the case of the Andaman Coucal Centropus andamanensis. From being a subspecies of the Brown Coucal Centropus sinensis (Ali and Ripley

1969), it is now elevated to a full species status, *Centropus andamanensis* by Rasmussen and Anderton (2005). With the change in taxonomic status, we have to evaluate the conservation status, mainly due to the small distributional range. It is also reported from the Coco and Table islands, and Myanmar. Rasmussen and Anderton (2005) have indicated that there is a "need for a comprehensive revision of *C. sinensis*." Perhaps, taxonomy based on DNA will be able to solve such problems.

The All Birds Barcoding Initiative (ABBI) is an international effort that aims to establish a public reference library of DNA barcodes for approximately 10,000 known bird species. The ABBI library of avian sequences linked to museum specimens will speed up discovery of new species and aid in the conservation of biodiversity. Approximately 7,000 individuals from 1,500 species have been barcoded so far. The BNHS, with its collection of 29,000 bird specimens, and the Zoological Survey of India, with its vast collection, can play an important role in barcoding Indian bird species.

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